REVIEW ARTICLE

Defect Reperfusion Imaging, a Newly Developed Novel Technology using Sonazoid in the Treatment of Hepatocellular Carcinoma

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The basic concept of contrast-enhanced ultrasonography has been confined to liver nodules, which are clearly depicted by B-mode ultrasound. However, by changing the classic concept of this method, a new breakthrough technique has been invented, using two favorable properties of Sonazoid: real-time vascular imaging and stable Kupffer phase imaging lasting up to 60 minutes. This new innovative technology called defect reperfusion imaging makes it possible to depict B-mode ill-defined nodules, locally recurring nodules, nodules found on screening/surveillance, and nodules found during staging. This is a major breakthrough technology, which will change clinical practice in hepatocellular carcinoma.

KEY WORDS — contrast-enhanced harmonic ultrasound, hepatic tumors, Kupffer phase imaging, radiofrequency ablation, treatment-assist guidance

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Introduction

The roles of various imaging techniques in the treatment of hepatocellular carcinoma (HCC) are shown in Table 1. Despite advances in diagnostic imaging techniques such as ultrasound (US), computed tomography (CT) and magnetic resonance imaging (MRI), there are still many difficulties related to HCC management, such as screening, staging, evaluation of treatment response, treatment guidance, localization of local recurrence after treatment, and diagnosis of recurrence after treatment (Table 2). Levovist-enhanced US has made a contribution to the differential diagnosis of HCC [1–3], as well as the evaluation of malignancy grade [4], evaluation of therapeutic response to transcatheter arterial chemoembolization [5–7], and needle insertion guidance [8,9]. However, there are still major limitations in the evaluation of therapeutic response to radiofrequency ablation (RFA) [10], as well as screening and staging of HCC.

The use of Sonazoid facilitates stable Kupffer phase imaging 10–60 minutes following injection and allows the acquisition of real-time blood flow

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images at low acoustic power (Table 3). Prior to the introduction of Sonazoid in this new technology, Sonazoid was considered to be more effective than Levovist in vascular imaging, was easy to use, and allowed visualization, even when using unsophisticated machines. Thus, dependence on skills/machines is decreased, which would facilitate the widespread use of contrast-enhanced US (Table 3). In addition, although Sonazoid US provides very stable postvascular phase images, its main purpose is considered to be the staging of metastatic liver cancer. Taking this into account, it was not expected that Sonazoid would markedly change clinical practice in HCC. However, this preconception has largely been rejected since its launch on January 10, 2007. In particular, since this epoch-making method was introduced, we have become convinced that Sonazoid US is an innovative breakthrough technology that will greatly change HCC daily practice. Defect reperfusion imaging involves Sonazoid reinjection into areas, which show defects in the postvascular phase [11]. The introduction of this method has led to dramatic improvements in the treatment problems associated with HCC, as shown in Table 2.

Table 1. Role of imaging in the management of hepatocellular carcinoma

Diagnosis
Screening (lesion detection)
Characterization (differential diagnosis)
Evaluation of malignant grade
Staging
Treatment
Evaluation of treatment response after transcatheter arterial chemoembolization/radiofrequency ablation
Treatment guidance
Localization of viable lesion after radiofrequency ablation
Detection of recurrence after treatment

Table 2. Problems which need to be solved in the management of hepatocellular carcinoma

Screening	Some tumors cannot be depicted by B-mode ultrasound due to coarse liver parenchyma
Staging	Difficult by B-mode or contrast-enhanced ultrasound (using Levovist or SonoVue)
Evaluation of treatment response	Computed tomography is frequently used after radiofrequency ablation Difficult to determine the safety margin if lipiodol is not injected
Localization of the local recurrence nodule after radiofrequency ablation	Difficult by contrast-enhanced ultrasound Not possible by B-mode ultrasound alone
Treatment guidance	Difficult for nodules which are not demonstrated by B-mode ultrasound
Detection of recurrence	Multi-detector row computed tomography every 3 months (cost, X-ray exposure, etc.)

Table 3. Comparison of contrast-enhanced ultrasound using Levovist and Sonazoid

	Levovist	Sonazoid
Microbubble	Air	Perfluorobutane
Shell	None	Yes (lipid)
Imaging technique	High mechanical index	Low mechanical index
Signal	Disruption (> harmonic)	Harmonic
Tumor vessel (angioarchitecture)	Real-time	Real-time
Tumor stain	Intermittent	Real-time
Kupffer phase (postvascular)	One sweep (one chance)	Stable for long period 10–60 minutes tolerable for multiple scanning
Operator-dependent	Yes	No or low
Machine-dependent	Yes	No or low

Development of Defect Reperfusion Imaging (Dual-phase Fusion Imaging)

We recently developed defect reperfusion imaging using the properties of very stable Kupffer images and real-time fine blood flow images obtained with Sonazoid for typical HCC, which can be depicted by CT but not by B-mode scanning. This is a breakthrough method for accurate localization and treatment guidance [11]. Diagnosis in dynamic studies is usually based on enhancing patterns according to time sequence or phase (Fig. 1). However, by changing the way of thinking, combined Kupffer and arterial phase images are obtained by Sonazoid reinjection (Fig. 2). This method is referred to as defect reperfusion imaging (Fig. 3).

Following intravenous injection of Sonazoid (0.01 mL/kg), early staining is observed in the vascular phase, and the presence or absence of defects can be determined in the Kupffer phase 10–60 minutes after injection. The probe is subsequently applied to the area, which shows a defect in the Kupffer phase. Sonazoid (0.01 mL/kg) is reinjected, and the presence of arterial blood flow entering the defective area is determined (defect reinjection method). When the presence of arterial vascularity in the Kupffer defect is regarded as a sign of reperfusion, the detection rate for typical HCC is almost 100% (Fig. 3). In addition, for nodules which cannot

Table 4. Ultrasound contrast agent

Contrast agent	Image	Approved country
Levovist	Vascular + Kupffer	Worldwide
SonoView	Only vascular	Europe, China, etc.
Definity	Only vascular	Canada, USA
Sonazoid	Vascular + Kupffer	Only Japan

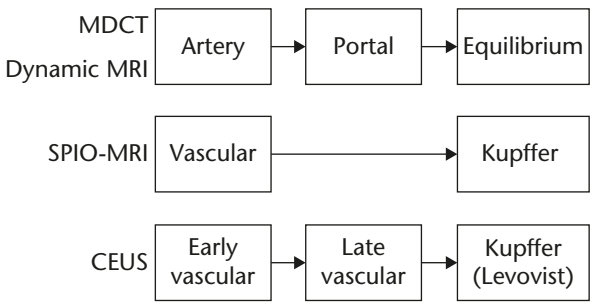

Fig. 1. *Normally, diagnostic imaging is performed based on enhancing patterns according to time sequence or phase. CEUS = contrast-enhanced ultrasound; MDCT = multi-detector row computed tomography; MRI = magnetic resonance imaging; SPIO-MRI = superparamagnetic iron oxide magnetic resonance imaging.*

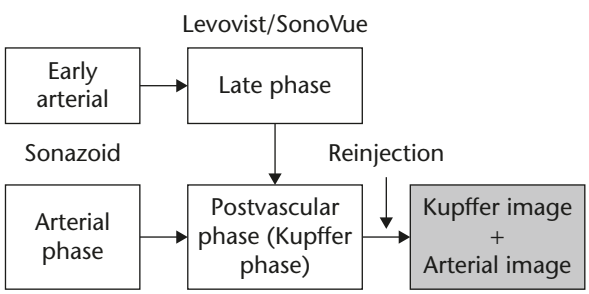

Fig. 2. *An innovative technique has been developed by reinjecting Sonazoid. Dual-phase imaging which combines Kupffer and arterial phase images is obtained by this method.*

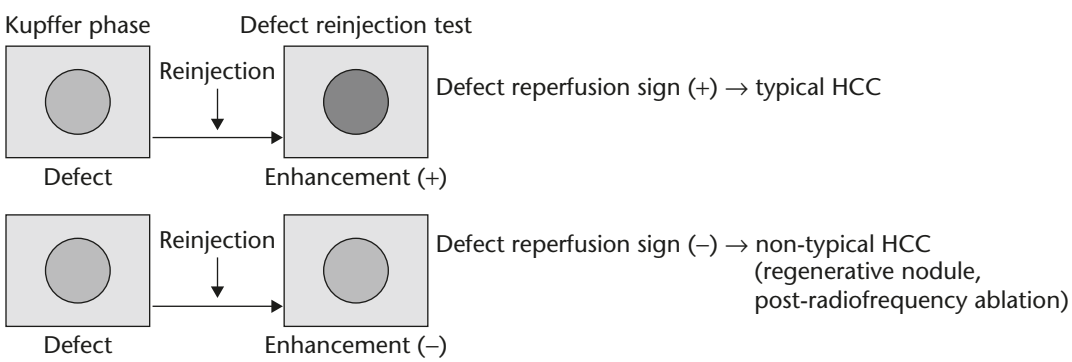

Fig. 3. *Even typical hepatocellular carcinoma (HCC) nodules that are not identified by B-mode ultrasound, which are shown as early enhancement with portal/venous washout on dynamic computed tomography, can be correctly diagnosed if arterial enhancement is obtained within the Kupffer defect area (Kupffer defect with hypervascularization).*

be visualized by B-mode US, Sonazoid can be reinjected in the postvascular phase where clear defects can be detected, but not in the vascular phase where arterial blood flow cannot be detected, and localization is not possible. Following the reinjection of Sonazoid, clear staining can be detected in the Kupffer defect (defect reperfusion sign, positive), and RFA, under the guidance of Sonazoid-enhanced US, is possible in cases where nodules are ill-defined by B-mode US, locally recurrent nodules or nodules found on screening (Fig. 4).

With this method, nodules which cannot be visualized by B-mode US are detected as defects on Kupffer images in the stable first Kupffer phase, and whether these nodules with Kupffer defects have arterial blood flow is subsequently determined by the reinjection test. This method may be a breakthrough in diagnostic imaging (Fig. 2) [11]. This contrast-enhanced US technique requires no special apparatus or analysis, and is the result of a change in the concept of contrast-enhanced US. For the typical CT image (so-called early enhancement with late washout nodules), defects are detected in the first Kupffer phase, and subsequently, perfusion of the defects is demonstrated by the reinjection test (visualization of staining within the Kupffer defects, which is the reverse phenomenon of early enhancement with late washout) (Fig. 3). The introduction of this innovative technique has allowed almost 100% identification of lesions which are shown on typical CT images but are not visualized on B-mode US images. If the reperfusion test showed no enhancement of a Kupffer defect, then this defect would differ from the nodule detected by CT (Fig. 3). Thus, this method may be an epoch-making treatment aid for HCC, which facilitates needle insertion guidance. In addition, defect reperfusion imaging has many possibilities, and is applicable in the screening of HCC in coarse liver parenchyma, localization of local recurrence after treatment, and punctures under the guidance of contrast-enhanced US (Fig. 4).

Application of Defect Reperfusion Imaging to the Screening, Staging and Treatment of HCC

Screening (depiction of HCC)

During screening, patients at high risk of HCC (type B and C liver cirrhosis) receive Sonazoid

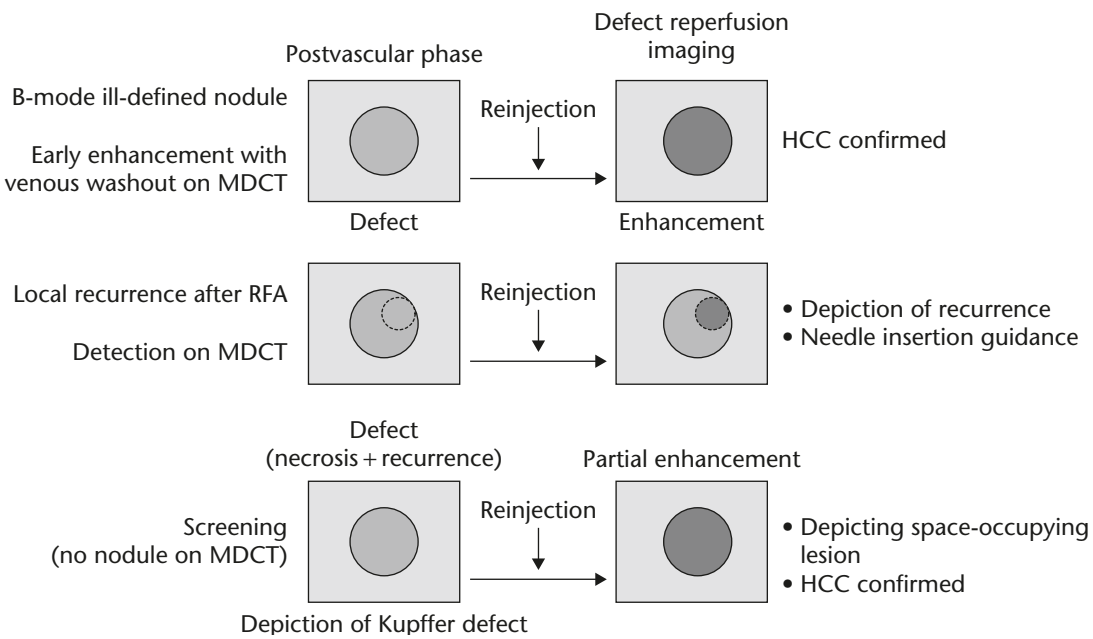

Fig. 4. *Defect reperfusion imaging is useful in the detection of nodules which are ill-defined by B-mode ultrasound, in addition to locally recurrent nodules and nodules depicted at screening/surveillance. HCC = hepatocellular carcinoma; MDCT = multi-detector row computed tomography; RFA = radiofrequency ablation.*

(0.01 mL/kg) by intravenous injection at an outpatient clinic. These high-risk patients then walk slowly to the US room where technologists perform Kupffer phase imaging between 10 and 60 minutes after Sonazoid administration. This procedure facilitates screening, because Sonazoid US can be performed in routine examination settings, without taking extra time for a contrast US study. In addition, operators need only concentrate on the depiction of Kupffer defects in the postvascular phase. If defects are detected, Sonazoid reinjection leads to the confirmation of HCC via information on both Kupffer function and arterial blood flow on the same cross-sectional image (Figs. 4 and 5). This dual-phase fusion imaging allows detection and definitive diagnosis of HCC with almost 100% confidence. In this respect, Sonazoid may also markedly improve the efficiency of HCC depiction during screening. Conventionally, contrast-enhanced US had been considered only for nodules previously depicted by B-mode US and was considered inappropriate for screening. However, this concept has been markedly changed with the innovation of defect reperfusion imaging using Sonazoid.

Staging

With the use of defect reperfusion imaging and Sonazoid, another breakthrough has been achieved in staging. Until now, CT during hepatic arteriography and CT during arterial portography have been the most sensitive staging tools. However, with Sonazoid-enhanced US, an entire liver scan can be performed in the postvascular phase, to precisely determine whether Kupffer defects are present in addition to the main nodule. To determine whether these Kupffer defects are intrahepatic metastases, the reinjection test is used [12], which allows accurate staging (Figs. 4 and 6). This is a huge advantage in the management of HCC.

Evaluation of treatment response

To date, the treatment effects of RFA have generally been evaluated by CT [12]. However, for the evaluation of the safety margin after RFA, a comparison of CT images before RFA is indispensable, and this comparison tends to be relatively inaccurate if lipiodol is not injected. On the other hand, the evaluation of treatment response using Levovist-enhanced US is practically impossible because of unclear nodule margins after RFA, the short enhancing period, or the fragility of Levovist. However, defect reperfusion imaging allows an accurate evaluation of the safety margin by visualization of the defects in the treated nodule in the postvascular phase, as well as determination of the ablation area, followed by determination of the safety margin using reinjection and cross-sectional scanning of the entire nodule (Figs. 7 and 8). Although further

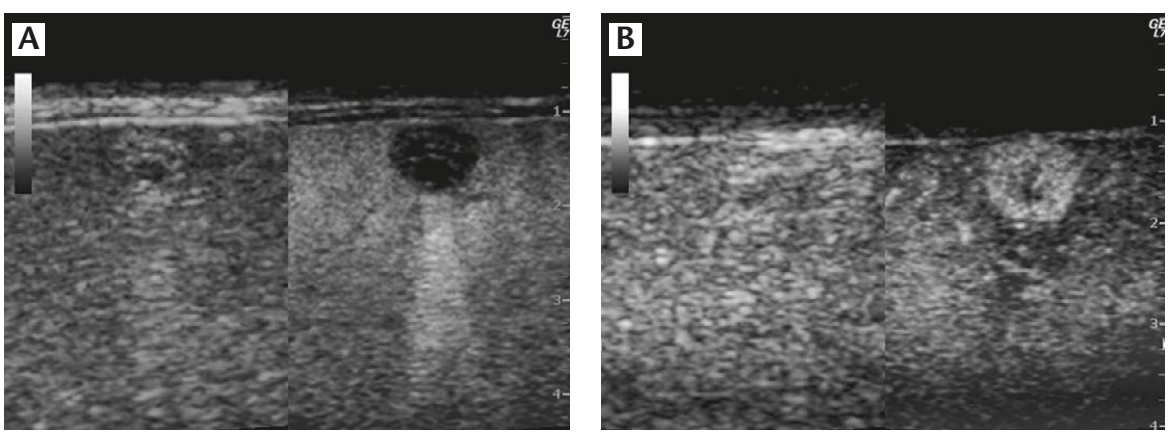

Fig. 5. *Hepatocellular carcinoma detected during screening of the entire liver. (A) Hepatocellular carcinoma detected during B-mode ultrasound (reference image) shows no clear nodules (left panel). However, Kupffer phase imaging at 11 minutes after intravenous injection of Sonazoid clearly shows Kupffer defects (right panel). (B) Reinjection of Sonazoid clearly shows arterial enhancement within the Kupffer defect (right panel).*

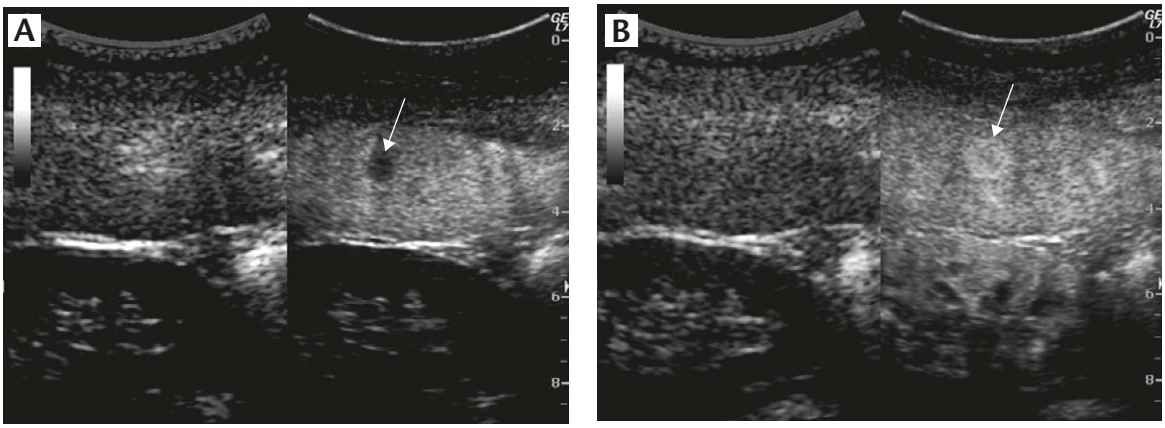

Fig. 6. *Staging of hepatocellular carcinoma. (A) Entire liver scan at Kupffer phase 16 minutes postinjection in a patient with a 3-cm hepatocellular carcinoma in S8 segment clearly showing a 5-mm Kupffer defect in segment 6 (arrow). (B) Reinjection of Sonazoid clearly shows this nodule to have arterial vascularization (arrow) with a Kupffer defect, which confirms that this nodule is an intrahepatic metastasis.*

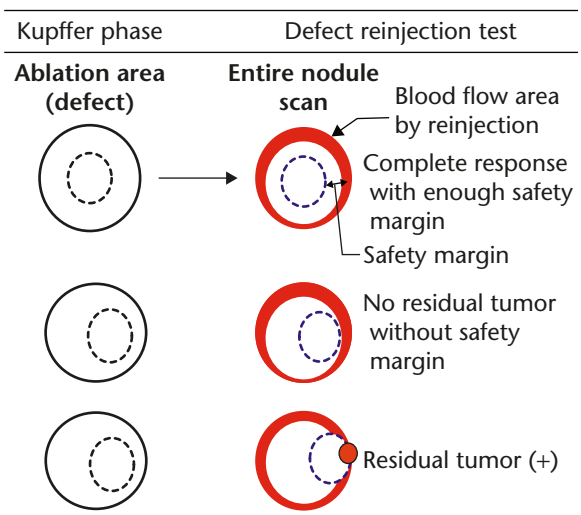

Fig. 7. *Evaluation of treatment response after radiofrequency ablation. It is possible to determine whether treatment response is complete with an adequate safety margin, residual tumor without sufficient safety margin, or positive staining.*

studies are necessary regarding the percentage of patients in whom the margin after treatment can be correctly determined, it is considered that CT can be omitted in approximately 50% of patients. The cost of each examination is ¥33,380 for CT but only ¥18,640 for Sonazoid-enhanced US. Thus, in the future, Sonazoid US may replace CT to some extent because of its advantages, in that it reduces cost and X-ray exposure and can also be performed in patients with contrast agent allergy or renal failure.

Visualization of locally recurrent lesions by US

Sonazoid is also useful for the localization of recurrent lesions located at previously ablated areas (because of the inhomogeneous echo pattern mixed with viable lesions, ablated area, and ablated surrounding liver). In this setting, even skilled operators have great difficulty in determining the viable area on B-mode US images alone, which correspond to the enhancing area on CT due to numerous US cross-sections [13]. This problem is readily overcome by defect reperfusion imaging with Sonazoid (Fig. 4).

Treatment guidance

Defect reperfusion imaging is particularly useful for needle insertion guidance in the treatment of HCC. For invisible nodules on B-mode US, needle insertion has been performed under the guidance of either real-time virtual sonography [12] or Levovist-enhanced US [8,9]. However, real-time virtual sonography requires CT volume data and special apparatus. In addition, complete concordance of synchronized images from B-mode US which correspond to the cross-sectional plane of CT volume data is sometimes difficult. Similarly, under Levovist-enhanced US, punctures should be performed within a very short time in the early vascular phase [7,8]. Because this is very difficult, this method is not widely accepted.

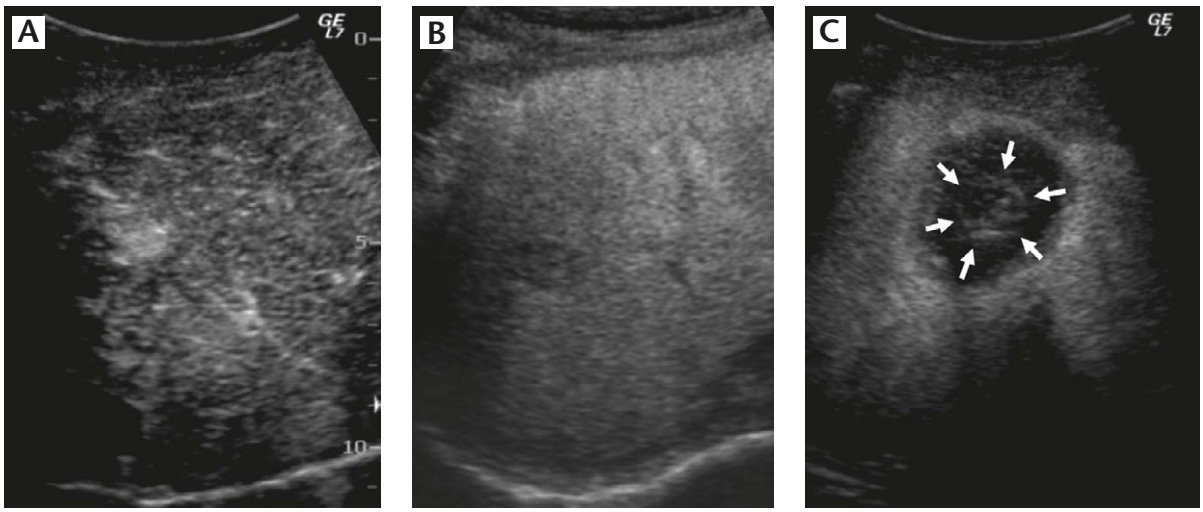

Fig. 8. *Evaluation of treatment response after radiofrequency ablation. (A) Hyperperfusion nodule is evident in segment 5. (B) Kupffer defect is evident in the postvascular phase. (C) Entire nodule scan by defect reperfusion imaging clearly shows a complete response with sufficient safety margin (arrows indicate original nodule).*

On the other hand, in Sonazoid-enhanced US, Kupffer defects can be detected very easily, and the presence of blood flow in defective areas can be determined by the reinjection method in all cases. Therefore, needle insertion can easily be performed during a stable period in the postvascular phase, and accurate needle placement followed by sufficient treatment is possible with Sonazoid-enhanced US (Fig. 4).

Diagnosis of distant recurrence after RFA

The diagnosis of local recurrence and recurrence in other areas is 100% possible using defect reperfusion imaging. Therefore, CT which is performed at 3-monthly intervals may be replaced by Sonazoid-enhanced US to some extent. This may also reduce costs and X-ray exposure.

Summary and Conclusions

Sonazoid-enhanced US is extremely useful in the characterization of hepatic tumors when compared with multi-detector row computed tomography or Levovist-enhanced US. More importantly, we are convinced that Sonazoid-enhanced US with defect reperfusion imaging is a new innovative approach to the treatment of HCC. This technique was invented, based on the two major favorable properties of Sonazoid: real-time blood flow images due to low acoustic power, and stable Kupffer images in the postvascular phase. Therefore, this breakthrough technology will markedly change the therapeutic strategy in liver cancer. Importantly, this technique is not possible with SonoVue or Definity (Table 4), which are approved in Europe, Canada and Asian countries including China and Korea, but only possible with Sonazoid. In this respect, Sonazoid should be made more available worldwide.

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